NOTES ON A NOVEL ABAXIAL LEAF EPIDERMIS IN ECUADORIAN BEGONIA PARVIFLORA

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Scanning-electron-microscope observations and stomatal-density counts indicate that two populations of Ecuadorian *Begonia parviflora* have very different surface epidermal characteristics. Compared to a population near Baños, Ecuador, a population near Tena has novel epidermal characteristics, including a mean stomatal density of 634.7 ± 26.2 stomata per mm^2, guard cells raised on cellular protrusions above the surrounding epidermal cells, and epidermal cells appearing indistinguishable from subsidiary cells. High stomatal density may be of adaptive significance to higher light, or exposure, intensity, although the possibility exists that raised guard cells and comparatively small epidermal cells are an architectural consequence of high stomatal density originating during early leaf development.

The genus *Begonia* L. is recognized for its unusual leaf anatomy, with many species having a multilayered epidermis (Fellerer, 1892; Solereder, 1908; Haberlandt, 1909; Metcalfe & Chalk, 1950; Foster & Gifford, 1959; Esau, 1965). Numerous species of *Begonia* have stomatal clusters, a characteristic limited to species in only a few unrelated angiosperm families (Fellerer, 1892; Metcalfe & Chalk, 1950; Neubauer, 1967; Boghdan & Barkley, 1972; Skog, 1976; Hoover, 1986). By contrast, other *Begonia* species have singly occurring stomata as well, which is the pattern exhibited by *B. parviflora* Poep. & Endl. This species is distributed at lower elevations on both the Amazonian and Pacific slopes of the Andes from Colombia to Peru. It is distinct from most species in the genus because it is a small tree (Smith & Schubert, 1941, 1946; Smith & Wasshausen, 1979). One of the most common Andean species of *Begonia, B. parviflora* is observed frequently as individuals or small populations and occasionally in colonies comprising many individuals.

In this study two populations of *Begonia parviflora* are compared for characters of the abaxial leaf epidermis. Although the populations grow in the same region, they differ considerably in density of stomata, length of stomatal pores, structural anatomy of guard cells, and size of epidermal cells.

METHODS AND MATERIALS

On February 3, 1984, two colonies of *Begonia parviflora* were sampled on the Amazonian slope of the Andes in Ecuador. The species was frequently

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observed throughout this region, with the colonies sampled being among the largest observed. The Baños colony was growing in moderate shade with successional vegetation 12 km east of Baños, Tungurahua Province (lat. 01°14'N, long. 78°22'W), at 1548 m altitude. The Tena population was observed at an exposed site 59 km north of Puyo, Napo Province (lat. 01°14'N, long. 77°53'W), at 906 m altitude. One leaf from each of ten healthy individual plants was collected at each population site, and clear fingernail polish was immediately applied to the lower leaf surfaces, allowed to dry thoroughly, then peeled off with forceps (Sampson, 1961).

In the laboratory the epidermal replicas were projected on a screen using an Edmund Scientific microprojector. Stomatal counts and measurements were made on a single mm^2 area from each peel. Lengths of five stomatal pores were measured on each peel and the mean determined. One clear epidermal replica was chosen from each population for permanent fixation for scanning-electron-microscope (SEM) observations. Preparatory methods for the SEM included placing the peels on aluminum stubs, situating the stubs with the peels in a vacuum, and coating the entire peel with a layer of gold about 150 Å thick. The specimens were then placed in the chamber of a Cambridge Stereoscan 100 SEM, observed from 300 to 800 ×, and microphotographed at about 300 ×.

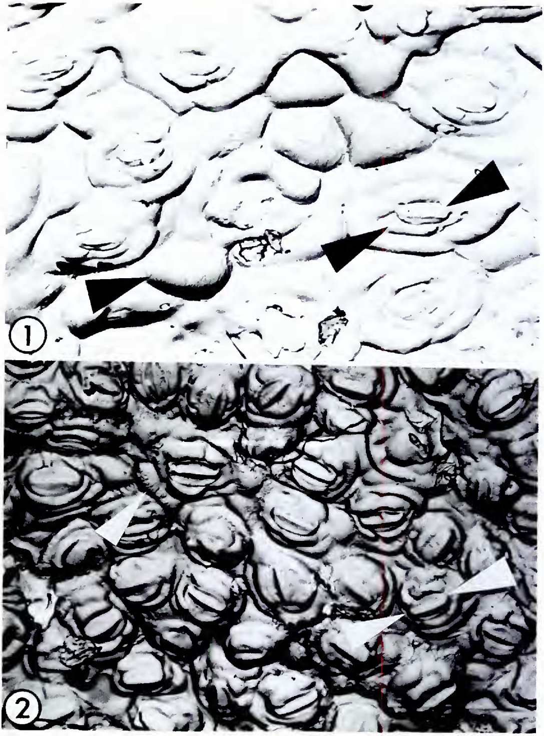
RESULTS AND DISCUSSION

The mean stomatal density for the Baños population of *Begonia parviflora* is 179.5 ± 19.13 per mm^2, and that for the Tena population is 634.7 ± 26.20 per mm^2 (see TABLE). Data were analyzed using a t-test at $P \leq .01$: $T_{18} = 44.4$. FIGURES 1 and 2 illustrate the difference between the two populations. The stomatal densities for the Tena population are among the highest reported for angiosperms (Salisbury, 1927; Carpenter & Smith, 1975; Abrams, 1987). In the literature available, species reported as having higher mean stomatal density values include (in stomata per mm^2) *Quercus muehlenbergii* Englem. (986), *Q. coccinea* Muenchh. (760), *Rhus copallina* L. (731), *Acer rubrum* L. (705), *Fagus grandifolia* Ehrh. (693), *Q. rubra* L. (681), *Ligustrum vulgare* L. (680), *Q. prinus* L. (662), and *Q. sessiliflora* Salisb. (656). Stomatal-density measurements from other species of *Begonia* are very limited, although several populations each of *B. heracleifolia* Schldl. & Cham. and *B. nelumbiifolia* Schldl. & Cham. have been sampled and found to have between 50 and 90 stomata per mm^2 (Hoover, 1986).

Ranges and means for stomatal density and pore lengths for B. parviflora populations.

	NUMBER OF STOMATA/MM²		STOMATAL-PORE LENGTH (in μm)	
LOCATION/*n*	Range	Mean	Range	Mean
Baños/10	91.7–280.0	179.5 ± 19.13*	8.0–15.0	10.4 ± 0.47
Tena/10	530.0–741.8	634.7 ± 26.20	6.0–22.0	8.4 ± 0.21

*Variance is measured as the standard error of the mean.



FIGURES 1, 2. Scanning electron micrographs of *Begonia parviflora,* abaxial epidermal surfaces: 1, Baños population, × 330; 2, Puyo population, × 405. (Arrows, left to right: epidermal cell, subsidiary cell, guard cell (raised in FIGURE 2)).

The stomata of the leaves from the Tena population seem to be raised above the plane of the epidermal cells (see FIGURE 2), and the guard cells appear to sit on the apex of individual cellular protrusions that are built up on subsidiary cells. Raised stomata have been observed in other species of *Begonia,* including

B. cathayana Hemsley, *B. froebelii* A. DC., *B. lobulata* A. DC., and *B. vitifolia* Schott (Brouillet, unpublished data). In general this is an uncommon characteristic in the plant kingdom, although it has also been reported in several other genera including *Pastinaca* L., *Prunus* L., and *Solanum* L. (Esau, 1965). The stomata of the leaves from the Baños population do not appear to have this structural modification; here subsidiary cells surround guard cells in the typical way (FIGURE 1).

In the leaves from the Tena population, compared to those from the Baños population, the epidermal cells are nearly indistinguishable from the subsidiary cells (see FIGURES 1, 2); they are greatly reduced in size and appear to be localized to depressions between the stomata. The stomata, with their associated subsidiary cells, are so densely packed that the epidermal cells on the surface are the same size as the subsidiary cells.

The mean stomatal length is 10.4 ± .47 μm for the leaves of the Baños population and 8.4 ± .21 μm for those of the Tena population, a statistically significant difference ($T_{18} = 3.83$ ($p \leq .01$)). Length of the stomatal pores has been measured in many different species of *Begonia* grown under cultivation, with results indicating relatively little interspecific variation in this character (Fellerer, 1892). Five different population samplings of *B. nelumbiifolia* from along an elevational gradient in Hidalgo, Mexico, indicate rather significant variation in pore length (Hoover, 1986), suggesting that this character may not be as stable as previously reported.

Rainfall along the Amazonian slope of the Andes is one of the few climatic parameters measured for this region (Schwerdtfeger, 1976). The highest annual rainfall recorded here is found at Tena, Ecuador (annual average of 6235 mm from 1965 to 1969, including the years 1968 and 1969, when accumulation totaled 8380 and 8939 mm, respectively), thus subjecting the *Begonia parviflora* population from this region to extreme moisture. Such high rainfall is unusual for the entire eastern region of the Andes, including Venezuela, Colombia, Peru, and Bolivia. At Puyo, 59 km south of Tena, rainfall averaged 4294 mm for the same years. No data are available for the region near the Baños population, although according to Schwerdtfeger (1976 p. 154) "rainfall distribution is highly variable, mainly because of the impact of relief." It is important to note that precipitation generally decreases with elevation in this region, and since Baños is 642 m higher than Tena, it likely receives less moisture and may experience somewhat of a dry season.

With only two populations sampled for *Begonia parviflora,* it is highly speculative to interpret these data from an evolutionary standpoint, even though the populations show such extreme morphological differences that they appear to represent different taxa. Limited data indicate that stomatal density increases when plants grow in exposed, sunny conditions (Jackson, 1967; Abrams, 1987, 1988). For these *B. parviflora* populations, the high-stomatal-density Tena plants were growing in an exposed, sunny habitat, while the low-stomatal-density Baños individuals grew under moderately shady conditions, thus suggesting that the high stomatal density may be of adaptive value to the exposed conditions. Although such an interpretation conforms with existing evidence, under the circumstances this interpretation may not be entirely adequate to explain the extreme morphological difference between the populations. Accommodation on average, of over 600 stomata per mm^2, including subsidiary cells, would predictably cause compression and reduction of epidermal cells, raising guard cells above the plane of the epidermal cells and reducing pore length. It would appear that these unusual morphological characteristics may be an architectural consequence of high stomatal density resulting during early leaf development, a process in accord with Gould and Lewontin (1978), who believed that morphological evolution may, in certain instances, be determined by structural change during development. Additional expeditions are planned for Ecuador and Colombia, data from which will assist in more thoroughly describing stomatal variation in *B. parviflora* and perhaps helping clarify the evolutionary implications of this novel leaf epidermis.

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LITERATURE CITED

ABRAMS, M. D. 1987. Leaf structural and photosynthetic pigment characteristics of three gallery-forest hardwood species in Northeast Kansas. Forest Ecol. & Managem. **22:** 261–266.

———. 1988. Genetic variation in leaf morphology and plant and tissue water relations during drought in *Cercis canadensis* L. Forest Sci. **34:** 200–207.

BOGHDAN, K. S., & F. A. BARKLEY. 1972. Stomatal patterns in the genus *Begonia.* Phytologia **23:** 327–333.

CARPENTER, S. B., & N. D. SMITH. 1975. Stomatal distribution and size in southern Appalachian hardwoods. Canad. J. Bot. **53:** 1153–1156.

ESAU, K. 1965. Plant anatomy. J. Wiley and Sons, Inc., New York.

FELLERER, C. 1892. Anatomie und Systematik der Begoniaceen. Unpubl. Ph.D. thesis, University of Munich, West Germany.

FOSTER, A. S., & E. M. GIFFORD, JR. 1959. Comparative morphology of vascular plants. W. H. Freeman and Co., San Francisco.

GOULD, S. J., & R. C. LEWONTIN. 1978. The spandrels of San Marco and the Panglossian paradigm: a critique of the adaptationist programme. Proc. Roy. Soc. London **205:** 581–598.

HABERLANDT, G. 1909. Physiologische Pflanzenanatomie. ed. 4. Wilhelm Engelmann, Leipzig.

HOOVER, W. S. 1986. Stomata and stomatal clusters in *Begonia*: ecological response in two Mexican species. Biotropica **18:** 16–21.

JACKSON, L. W. R. 1967. Effect of shade on leaf surfaces of deciduous tree species. Ecology **48:** 498, 499.

METCALFE, C. R., & L. CHALK. 1950. Anatomy of dicotyledons. Clarendon Press, Oxford.

NEUBAUER, H. F. 1967. Bemerkungen über den Bau der Begoniaceen. Ber. Deutsch. Bot. Ges. **80:** 80–97.

SALISBURY, E. J. 1927. On the causes and ecological significance of stomatal frequency with special reference to the woodland flora. Philos. Trans., Ser. B. **216:** 1–65.

SAMPSON, J. 1961. A method of replicating dry or moist surfaces for examination by light microscopy. Nature **191:** 932, 933.

SCHWERDTFEGER, W. 1976. Climates of Central and South America. World survey of climatology. Vol. 12. Elsevier Scientific Publishing Co., Amsterdam.

SKOG, L. E. 1976. A study of the Gesnerieae, with a revision of *Gesneria* (Gesneriaceae: Gesnerioideae). Smithsonian Contr. Bot. **29:** 1–182.

SMITH, L. B., & B. G. SCHUBERT. 1941. Flora of Peru: *Begonia.* Field Mus. Nat. Hist. Bot. Ser. **13:** 182–202.

——— & ———. 1946. The Begoniaceae of Colombia. Caldasia **4:** 77–107.

——— & D. C. WASSHAUSEN. 1979. *Begonia* of Ecuador. Phytologia **44:** 233–256.

SOLEREDER, H. 1908. Systemic anatomy of the dicotyledons. Vols. 1, 2. Clarendon Press, Oxford.